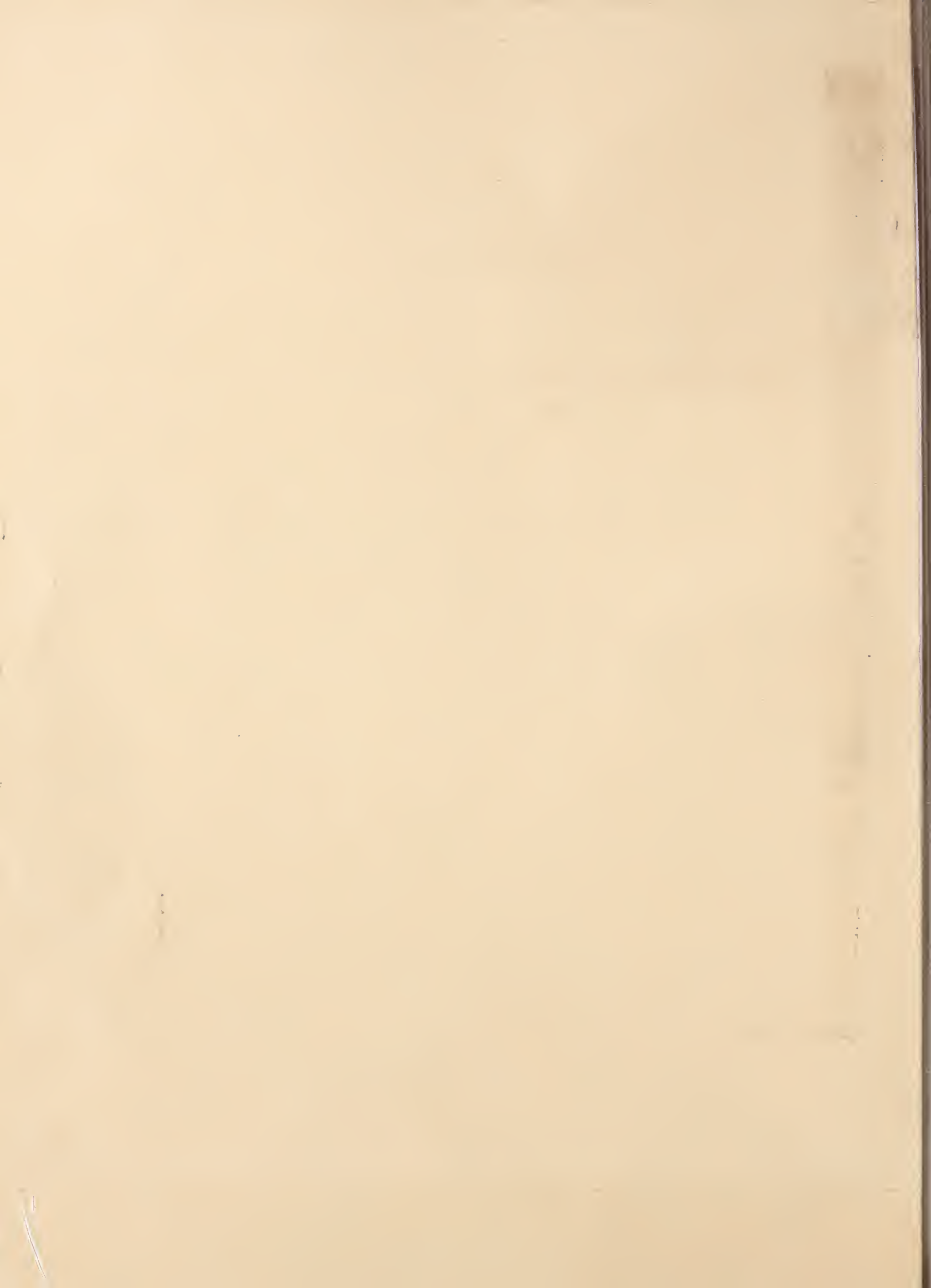


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DOUGLAS-FIR

BARK TANNIN DECOMPOSITION IN TWO FOREST SOILS

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INTRODUCTION

About 5 million tons of tree bark are produced annually as waste from wood-using industries in the Pacific States. Douglas-fir is the principal species involved. Most of this bark is burned, a practice which contributes significantly to air pollution in areas of mill concentration. Alternative methods of disposing of tree bark include agricultural or horticultural use as mulches or soil conditioners. Another possibility is to debark trees at the logging site and return the bark to the soil for stabilization and organic matter maintenance.

Known effects of adding bark to soil have been summarized (Bollen 1969),^{1/} but one aspect of the problem has not hitherto been closely examined--the effect of bark tannins on the soil.

Tree barks contain various amounts of tannins, water soluble polyphenols capable of forming complexes with proteins and other organic substances. Douglas-fir bark contains from 5 to 22 percent tannin by weight (table 1), depending on tree age and section of the stem from which the bark came (Kurth et al. 1948). Bark has been applied to soil at rates of up to 100 tons per acre, so as much as 20 tons per acre (20,000 parts per million (p.p.m.) per acre 2 million pounds of soil) of tannins might thus be added. Bark residues on forest and cut-over lands also add tannins.

To answer questions concerning the fate of these bark tannins in soil and their effect on the soil, we posed these questions: (1) How rapidly do tannins decompose in soil? (2) Do they affect microbial populations and accumulation of nitrate nitrogen at a concentration possible from common applications of bark mulches and incorporations in agriculture?

Barks of other western tree species also contain tannins in amounts sufficient to require consideration when bark is added to soil (table 1).

REVIEW of PREVIOUS RESEARCH

Certain plant pathogenic fungi are inhibited and finally killed by tannin added to culture media, but saprophytic fungi are less sensitive (Cook 1911). *Aspergillus niger*, a common soil mold, can thrive on tannin and has been used commercially to hydrolyze tannin to gallic acid (Prescott and Dunn 1959). Koch and Oelsner (1916) reported that tannin is readily assimilated by soil molds. Certain cellulolytic bacteria are inhibited by tannins at 100 to 1,000 p.p.m., but growth

^{1/}Names and dates in parentheses refer to Literature Cited, p. 10.

Table 1.--Tannin in bark of some Pacific Northwest tree species

Species	Bark tannin	Reference
	<u>Percent</u>	
Douglas-fir (<i>Pseudotsuga menziesii</i>)	8-18 5-22	Kurth (1953) Hathway (1960)
Western hemlock (<i>Tsuga heterophylla</i>)	15-16	Howes (1953)
Sitka spruce (<i>Picea sitchensis</i>)	11-37	Frey and Clarke (1941)
Ponderosa pine (<i>Pinus ponderosa</i>)	6-11	Kurth and Hubbard (1951)
Sugar pine (<i>Pinus lambertiana</i>)	5-7	Kurth et al. (1949)
Western larch (<i>Larix occidentalis</i>)	16	Jarvis (1959)
Western redcedar (<i>Thuja plicata</i>)	5-7	Smith and Kurth (1953)
Incense-cedar (<i>Libocedrus decurrens</i>)	4-8	Smith and Kurth (1953)
Redwood (<i>Sequoia sempervirens</i>)	1-3 2-8	Wilson and Thomas (1927) Gnamm (1949)
Port-Orford-cedar (<i>Chamaecyparis lawsoniana</i>)	4-7	Smith and Kurth (1953)
Mountain hemlock (<i>Tsuga mertensiana</i>)	16-21	Kurth (1958)
Red alder (<i>Alnus rubra</i>)	4	Kurth and Becker (1953)
Grand fir (<i>Abies grandis</i>)	7	Kurth and Tokos (1953)
Red fir (<i>Abies magnifica</i>)	16	Becker and Kurth (1958)
White fir (<i>Abies concolor</i>)	6-10	Hergert and Kurth (1953)

of others, except for changes in morphology, is only slightly affected (Henin, Tagari, and Volcani 1964). Shrikhande (1940) showed that fermentation of tea refuse was not impeded by the tannin complex, but its decomposition was qualitatively altered by selective action on the microflora.

Douglas-fir bark decomposes in soil more slowly than sawdust. The bark bast is more resistant than the cork fraction (Allison 1965; Bollen and Glennie 1961), which contains tannins, dihydroquercitin, and other more or less toxic extractives. Aspitar^{2/} treated Willamette silty clay loam soil with Douglas-fir tannin liquor equivalent to 2,000 p.p.m. carbon and found 33 percent of the tannin was decomposed in 60 days.

Basaraba and Starkey (1966), using carbon dioxide evolution as an index, showed that tannins combine with proteins and reduce the rate of decomposition of the complexes in liquid media inoculated with soil. However, chestnut (*Castanea dentata*) and wattle (*Acacia* spp.) tannins alone decomposed more slowly than the tannin-protein complexes.

Benoit, Starkey, and Basaraba (1968) found that wattle tannin reduced decomposition of whole plant materials about 50 percent in inoculated culture media having pH values of 4.0 and 7.0. Although protein-tannin complexes were resistant, the effect of tannin on the whole-plant materials was not correlated with their nitrogen content and was, therefore, believed due to causes other than the formation of complexes with plant proteins. Benoit and Starkey (1968) found that tannin reduces activity of cellulase and other exoenzymes of micro-organisms involved in decomposing organic compounds of large molecular weight, such as cellulose and proteins. They concluded that reduction of the exoenzyme activity is an important factor in the inhibitory effect of tannins on decomposition of plant residues. Tannins thus may influence the accumulation of organic matter, particularly in forest soils, and may also promote persistence of organic matter in other soils to which bark is applied.

Lewis and Starkey (1968) found that, in a hardwood forest soil, the hydrolyzable tannins, tannic acid, gallotannin, and chestnut tannin, as well as the simple tannin, catechin, underwent appreciable decomposition in 60 days as indicated by CO₂ evolution. Only a little CO₂ was released from tannins of wattle and canaigre (*Rumex hymenosepalus*). Resistance of tannin-protein complexes to decomposition varied with the tannins and proteins. The tannins also retarded decomposition of starch, chitin, saponin, and, to a lesser degree, pectin.

MATERIALS AND METHODS

Douglas-fir bark was first treated sequentially with hexane, benzene, ether, and 95 percent ethanol^{3/} to remove waxes, flavonoids, and phlobaphenes. After

^{2/}Aspitar, T. R. Availability of nitrogen in ammoniated bark used as a soil amendment. 1959. (Unpublished Ph.D. thesis on file at Oreg. State Univ., Corvallis.)

^{3/}By Dr. Harvey Aft, Forest Research Laboratory, Oregon State Univ., Corvallis.

this initial treatment, tannin was extracted from the bark with hot water. The dried extract contained 97 percent tannin and about 0.1 percent sugars. Analysis of the powdered extract gave the following results (percent): water 8.33; ash 2.07; carbon 55.69; nitrogen 0.12. The ash and nitrogen are extraneous, representing unavoidable natural contaminations of bark on the tree.

Two forest soils differing widely in organic matter content and microbial populations (table 2) were used. One, the A1 horizon of an Astoria-like silty clay loam hereafter referred to as "Astoria soil," was collected beneath a 40-year-old stand of mixed Douglas-fir, western hemlock, and Sitka spruce. The other was the C horizon of Preacher clay loam, beneath a Douglas-fir-western hemlock forest. The soils were air-dried and passed through a 10-mesh screen before use.

Two-hundred-gram portions, oven-dry basis, of each soil were treated in triplicate as follows: (1) soil only; (2) soil plus dextrose equivalent to 2,000 p.p.m. carbon; and (3) soil plus the purified tannin equivalent to 2,000 p.p.m. carbon or 3,775 p.p.m. (0.38 percent) tannin. This tannin addition approximates the amount of tannin leachable from a 2-inch bark mulch, if we assume a tannin content of 10 percent. A mulch this deep, common in actual use, weighs 20 tons per acre, oven-dry basis. The actual amount of tannin leached and its concentration in the underlying soil will vary with the amount and timing of precipitation or irrigation and with depth of penetration of water into the soil.

Microbial populations, nitrate content, and nitrification were determined by methods of Bollen et al. (1967). Carbon dioxide evolution was determined according to Bollen and Lu (1957).

RESULTS AND DISCUSSION

TANNIN DECOMPOSITION

Decomposition of native soil organic matter, tannin, and dextrose was measured as the amount of CO₂ evolved during incubation (Bollen and Lu 1957). Dextrose, a readily available source of carbon, served as a standard for comparison with other treatments because it is rapidly decomposed by microbes.

Native organic matter, dextrose, and tannin all decomposed at a greater rate in the highly organic Astoria soil than in the Preacher soil of very low organic matter content (fig. 1). Part of the CO₂ which evolved from soils to which tannin or dextrose had been added may have come from native soil organic matter. When a carbonaceous substance is added to soil, the heterotrophic microflora population is stimulated and microbial attack on native soil constituents increases (Hallam and Bartholomew 1953); this is the "priming effect" discussed by Starkey (1968). We show "apparent decomposition" in figure 1 to best indicate the real amount of tannin and dextrose decomposed. Apparent decomposition is expressed as the percentage of carbon evolved as CO₂ from native and added organic matter in treated soil less that given off by untreated soil.

Table 2.--*Chemical and physical properties of Astoria and Preacher soils*^{1/}

Chemical properties	Astoria clay loam (A1 horizon)	Preacher loam (C horizon)
Total carbon (percent)	14.48	0.24
Nitrogen (p.p.m. ^{2/}):		
NH_4^+	17.00	.00
NO_2^-	.10	.00
NO_3^-	21.00	1.1
Kjeldahl ^{3/}	7,400.00	200
Carbon: nitrogen ratio	19.00	12
pH	5.30	4.6
Total cation exchange capacity (meq. per 100 g. ^{4/}):	69.0	19.1
Exchangeable H^+	30.4	16.7
Exchangeable Ca^{++}	12.5	1.7
Exchangeable Mg^{++}	16.0	.4
Exchangeable K^+	5.2	.3
Sum of cations	64.1	19.1
Available phosphorus (p.p.m.)	4.30	3.0
Physical properties:	- - - - - Percent - - - - -	
Water-holding capacity	206	57
Water retention at:		
0.1 atmosphere	79.84	42.6
0.33 atmosphere	58.15	33.1
1.0 atmosphere	44.69	25.3
15.0 atmospheres	31.14	14.8
Mechanical analysis:		
Sand	28.9	42.35
Silt	47.1	39.64
Clay	23.9	18.01

^{1/} Analytical methods are described in Bollen et al. (1967).

^{2/} Parts per million.

^{3/} Includes NO_2^- -N and NO_3^- -N.

^{4/} Milliequivalents per 100 grams.

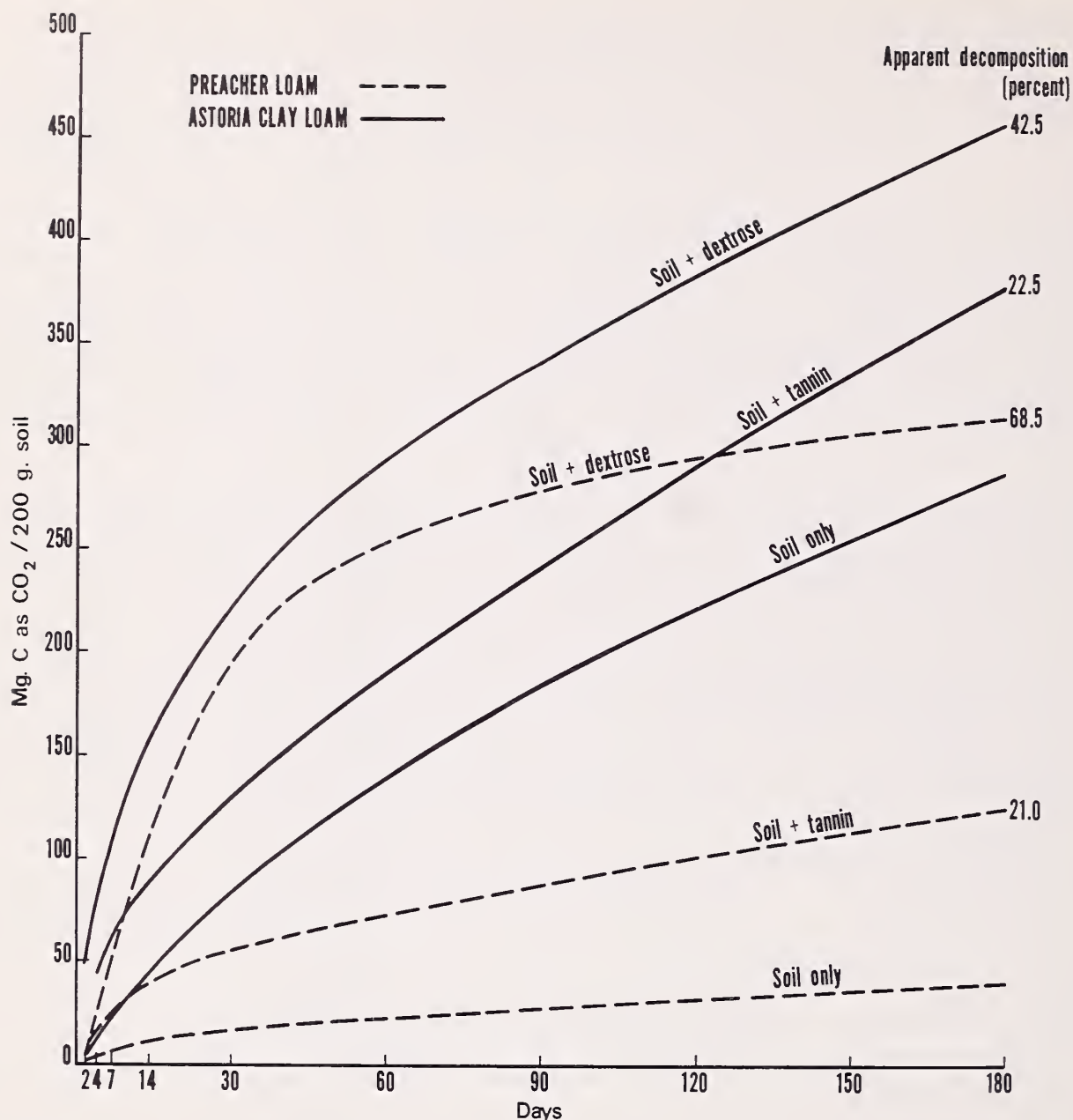


Figure 1.--Influence of Douglas-fir bark tannin and dextrose on CO₂ production. Preacher loam, and Astoria clay loam.

Total CO₂ evolution from Astoria soil plus dextrose was much higher than that from Preacher soil plus dextrose. However, untreated Preacher soil produced much less CO₂ than did the untreated Astoria soil, so apparent decomposition of dextrose in the Preacher soil was much higher, 68.5 percent, than in Astoria soil (42.5 percent). Total evolution of CO₂ from soil to which tannin had been added was also much greater in Astoria than in Preacher soil, but again the higher CO₂ production by Astoria soil alone compensated, in this case resulting in an apparent decomposition of tannin at about the same level in both soils--nearly 22 percent.

At least two additional factors further lessen the impact of tannin on soil properties. From 2 to 5 percent of bark residues from lumber mills is wood particles, the tannin content of which is negligible. In addition, bark contains an abundant microbial population which undoubtedly decomposes some of the tannin before it can leach into the soil.

MICROBIAL POPULATIONS

Koch and Oelsner's (1916) report that adding tannin to soil greatly favors development of molds is supported by our findings (table 3). Incubation of untreated soil substantially increased numbers of molds in both soils studied, but incubation of soil to which tannin was added further increased their numbers by 10 times in Astoria and 25 times in Preacher soil (table 3).

Some changes in predominant kinds of molds were also noted. In the Preacher soil alone, incubation increased *Penicillium* percentage, but incubation with tannin caused no further change in proportions. In the Astoria soil, incubation decreased percent of *Penicillium*, but incubation plus tannin caused a predominance of this species of mold. Other mold colonies, not identified, were almost exclusively compact, white, and sterile.

Table 3.--Microbial populations in soil--unincubated, incubated, and
incubated with tannin treatment
(Means of three replications)

PREACHER LOAM				
Treatment and days incubated	Molds		Bacteria	
	Total	<i>Penicillium</i>	Total	<i>Streptomyces</i>
	Thousands per gram	Percent	Millions per gram	Percent
Soil alone (0)	3	68	0.05	34
Soil alone (180)	43	94	2.00	0
Soil+tannin ^{1/} (180)	1,073	98	4.80	2
ASTORIA CLAY LOAM				
Soil alone (0)	195	39	9.30	45
Soil alone (180)	240	23	20.90	69
Soil+tannin ^{1/} (180)	2,472	93	42.20	92

^{1/} Tannin was added at the equivalent of 2,000 p.p.m. carbon.

Bacterial population was also affected by the treatments, but not so strikingly as were molds. Incubation markedly increased numbers of bacteria, especially in the Preacher soil, and incubation plus tannin caused a further doubling.

Streptomyces spp. were eliminated in incubated Preacher soil and also, to a large extent, in the incubated tannin-treated soil. In Astoria soil, on the other hand, incubation increased percentage of *Streptomyces* and incubation of tannin-treated soil resulted in an even greater preponderance of these higher bacteria.

It should be noted that bark carries a high microbial population (Bollen 1969) which undoubtedly decomposes some of the tannin before it can be leached into the soil. Thus, in a bark mulch, the same initial level of tannin as that used in our experiment would likely affect soil microbial activities less than did the extracted tannin concentration we used.

NITRIFICATION AND NITRATE PRODUCTION

In the low-nitrogen Preacher soil, nitrates in untreated samples increased from an initial 1.1 p.p.m. to 1.5 p.p.m. after incubation (table 4). However, the nitrate level in incubated soil to which tannin had been added was only 1.3 p.p.m. Results of nitrification studies with the Astoria soil were of greater magnitude because of higher beginning nitrogen content, and the decreases due to tannin addition were more pronounced. In this case, a definite, though not extensive, retardation of nitrification occurred.

In some areas increases in nitrate production after timber harvesting have been shown to pose a hazard to stream water quality. Bormann et al. (1968) found nitrate concentrations in a small stream draining a cutover ecosystem to have exceeded established pollution levels (10 p.p.m.) for more than 1 year after cutting, and algal blooms appeared during the summer period. Bollen (1969) suggested that tree bark might best be left on the forest soil after timber harvesting to provide physical protection against soil erosion, to allow some return of organic matter and mineral nutrient elements to the soil, and especially to aid in reducing undesirable buildup of leachable nitrates. Evidence from our study of tannin effects on nitrification appears to further support the desirability of leaving as much bark on the cutting area as possible in order to slow nitrification and thus reduce excessive formation of water-polluting nitrates.

SUMMARY

Addition of tannin to two greatly different forest soils produced no evident harmful effects. However, the moderate decrease in nitrification effected by tannin bears consideration in agricultural use. This feature could even be useful in watershed management because leaving bark on forest soils may lower nitrate production and thus decrease the hazard of nitrates in water supplies fed by forest streams.

Table 4.--*Nitrification of native organic matter and added tannin after incubating two soils*

(Means of three replications)

PREACHER LOAM			
Treatment and days incubated	NO ₂ -N	NO ₃ -N	Net NO ₂ -N plus NO ₃ -N after incubation
	----- P.p.m. -----		
Soil alone (0)	0	1.1	--
Soil alone (180):			
Gross	.02	1.5	--
Net	.02	.4	0.42
Soil+tannin ^{1/} (180):			
Gross	.10	1.3	--
Net	.10	.2	.30
ASTORIA CLAY LOAM			
Soil alone (0)	.10	21.0	--
Soil alone (180):			
Gross	.60	86.0	--
Net	.50	65.0	65.50
Soil+tannin ^{1/} (180):			
Gross	.10	77.0	--
Net	.00	56.0	56.00

^{1/} Tannin was added at the equivalent of 2,000 p.p.m. carbon.

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Purified Douglas-fir bark tannin equivalent to the amount in a 2-inch bark mulch was added to two widely different soils in the laboratory. About 22 percent of the tannin decomposed in 180 days. Soil microflora generally increased. Nitrate production was slightly decreased in the presence of tannins. Douglas-fir bark added to soil appears harmless from the standpoint of its tannin content.

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